ESSAY ON

THE CONDITIONS AFFECTING THE VISCOSITY OF THE BLOOD.

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by

M. H. FINKELSTEIN

from the Department of Bacteriology,

Edinburgh University.

PART I.

HISTORICAL INTRODUCTION.

The subject of the viscosity of the blood is of comparatively recent development. The experimental study of viscosities commenced with the work of Poiseuille on hydrodynamics in 1839. Prior to this date and indeed as far back as classical times we find many clinical references to "the thickness of blood", changes in which, appear to have impressed many early observers, thus, Ambrose Pare, the famous surgeon, commenting on asphyxia, observes that blood flowing from the wounds of the asphyxiated, is exceedingly "thick".

Mangetus gives a number of preparations, many of peculiar composition, which are supposed to render the blood more "watery".

C. 1839. Poiseuille. So far as the viscosity of the blood is concerned, Poiseuille's contribution consists of the formulation of the fundamental laws regarding the flows of fluid from a tube. The law referred to may be stated as follows:-

(1) The amount of outflow is proportional to the head of pressure.

(2) The time for outflow of a constant volume at constant/

constant pressure is directly proportional to the length of a tube of constant diameter.

(3) For a constant pressure the time for outflow of constant volume per tubes of constant length is inversely proportional to the fourth power of the diameter. Or, if A is volume outflow per second, r & l radius and length of the tube, p the pressure, and k the constant

$$A = \frac{k r^4 p}{l}$$

$$v = \frac{k r^2 p}{\pi l}$$

It is important to note that Poiseuille's law holds only for fluids in which there is orderly flow.

Poiseuille introduced a viscosimeter in which the liquid escapes from a reservoir through a horizontal capillary. The viscosity is obtained through the time taken to empty the reservoir.

Poiseuille himself carried out no essential investigations on the viscosity of the blood.

1860 Jacobson is responsible for most of the early work on the experimental confirmation of Poiseuille's Law.

1871 Duncan and Gamgee find that the viscosity of blood is 4 to 5 times greater than water and are the first to discover that defibrinated blood possesses a lower viscosity than unclotted blood.

1876 Haro is one of the first investigators to determine the viscosity of blood, which he finds to be in the case of man, about 4 times that of water. The viscosimeter used by this investigator was of the Poiseuille type.

1877 Ewald measures the viscosity of blood by an instrument of the Poiseuille type and finds it to be 4 to 5 times that of water. He also notes that a mixture of blood and water is less viscous than blood.

1896 Nicolls again working with a modification of the Poiseuille instrument so constructed as to be insertable into a large artery, determines the viscosity of blood, to be about 4 times that of water. His researches principally concern oxalated blood.

1897 Levy states that the viscosity of blood is 3½ times that of distilled water. His observations are carried out on defibrinated blood. This may account for the low figure which he obtained.

1900 Hürtle and Burton-Opitz introduces a new method for investigating viscosities, known as Hurtle's method. This consists in measuring the quantity of blood which flows per unit of time from a/

a capillary of known diameter inserted into a vessel. It has the great advantage of dealing with uncoagulated blood to which no anticoagulin has been added. In order that the pressure under which the flow takes place shall be known, the blood-pressure of the animal requires to be determined.

By means of this method the earlier results of Levy, Ewald and other workers are, in general, confirmed and it is found that oxalated or defibrinated blood has a lower viscosity than blood in the vessels.

1901 Hirsch and Beck are the first observers to use a method devised by Ostwald in which the blood is allowed to flow under gravity through a narrow capillary and the velocity of flow measured.

The viscosity of human blood they find to be 5 times that of water.

1902. The first investigations of Burton-Opitz on the relative viscosities of blood of different animals, show that the blood of the turtle and the frog have considerably less viscosity than the blood of mammalia.

1904. Using Hurtle's method Burton-Opitz shows that the effect of alcohol given intravenously or into the stomach raises the viscosity of blood as much as 20%.

1904 Müller demonstrates for the first time that the administration of Iodides lowers the viscosity of the blood. Great clinical significance has been attached to this finding.

1905 Burton-Opitz demonstrates that during narcosis with ether, chloroform or morphia, the viscosity of the blood does not alter significantly. This is an important finding for it allows many experiments to be done under anaesthesia without fallacies appearing in the results.

1905 Bence in his investigations on the effect of variation of the gaseous content of the blood, observes that CO_2 administration greatly increases the viscosity of blood. This effect is due to a change in the red-cells for the viscosity of serum is unaltered by CO_2 saturation. Similarly O_2 given to a dyspnoeic animal lessens the viscosity of the blood and may render it normal again.

1905 A remarkable claim is put forward by Faro and Rossi who state that the thyroids and para-thyroids furnish an internal secretion which regulates the viscosity of the blood. This claim has been adversely criticised by many observers and the results of their experiments have not been confirmed.

1906/

1906. Burton-Opitz working by Hurtle's method, observed the effect of hot and cold baths on the viscosity of the blood in man. Hot baths 43°C decrease the viscosity about 12% while cold baths increase the viscosity by about the same amount. Burton-Opitz points out that these changes have a relation to an alteration in the specific gravity which takes place in the blood, in hot and cold baths respectively. Hot-air baths act in the opposite way to hot-water baths. The former increase the viscosity and the specific gravity, whereas the latter decrease the viscosity and specific gravity.

1906 Burton-Opitz finds that hyperglycaemia induced by injection of glucose increases both viscosity and specific gravity.

1906 Burton-Opitz finds that in anaemia from haemorrhage the viscosity falls, sometimes as much as 20% or 30%, depending on amount of blood withdrawn. This is due to the diminished cell-content. It is interesting to observe that phenyl-hydrazine which also produces anaemia does not lower the viscosity. This, however, Burton-Opitz shows is due to the fact that large and irregular cells take the place of the normal erythrocytes.

1907 Heubner writes a critique of the different methods of estimating viscosity and criticises the /

the applications of Poiseuille's law to non-rigid tubes such as the capillaries in an animal. He concludes that the viscosity effects which occur in blood circulating through the capillaries bear no direct relation to the viscosity effects observed experimentally in vitro.

1907 Du Bois Reymond, Brodie and Müller by perfusing living organs show that Heubner's objections are invalid and that Poiseuille's law applies in vivo as in vitro.

1908 Burton-Opitz investigating the effects of CO_2 confirms Bence's observations (vide supra) and also shows that the viscosity of venous blood is slightly greater than arterial.

1908 Burton-Opitz arrives at the conclusion that the viscosity of blood depends entirely on the red-cell content, increasing as the red-cell content increases.

1908 Dettermann introduces his viscosimeter which consists of a capillary with bulbs at each end and which can be inverted. The time taken for the blood to flow between two marks on the capillary measures the viscosity. The instrument is calibrated with water. Dettermann contradicts Burton-Opitz's statement that the viscosity of blood laked by freezing and thawing is less than that of normal blood (1908) Burton/

Burton-Opitz in his turn attributes Dettermann's results to incomplete freezing but in a later paper (1908) attributes the difference between the two findings, to the presence or absence of the envelopes of the laked cells. If these are present the viscosity is increased. If they are centrifuged off as in Burton-Opitz's original investigations, the viscosity is decreased.

1920 Hess introduces his modification of the Couette method. This method depends upon the velocity of rotation of coaxial cylinders separated by the fluid whose viscosity is to be determined. The principle was first considered by Saint-Venant in 1847 and by Stokes in 1845 and elaborated by Boussinesq and by Margoules in 1873 and 1881 respectively. A practical method was devised by Couette in 1890 and a very much perfected instrument by de Nouy in 1923. A description of the instrument will be found in the section on method, so far attention has been principally directed to the perfection of the apparatus and the results of many of its possible applications are not yet forthcoming.

Besides the above, there are many references of a purely incidental nature, to the viscosity of blood. None of these are of first importance and have therefore been omitted. For example in the recent papers on sedimentation the viscosity of the blood or other fluids is referred to as an important factor (Ponder 1926).

It is of importance to correlate if possible, the investigations on the viscosity of blood, with investigations on the viscosity of emulsoids and suspensoids, since blood contains emulsoid systems and is when taken as a whole a suspensoid system.

Most of the physical observations have been made since 1905. For suspensoids the results of viscosity determinations vary greatly, being dependent principally on the volume of the suspended phase. The viscosity of dilute suspensoids has been studied by an enormous number of observers commencing with Friedlander and including Botazzi and Ostwald. The general result of these investigations is:-

(1) That very dilute suspensoids, e.g. glycogen solutions or silver sols possess viscosities scarcely different from that of water.

(2) That as the concentration of suspensoid phase increases, the viscosity increases as a linear function until a certain critical point.

(3) That after the critical point, increase of concentration is accompanied by an enormous increase of viscosity.

For emulsoids the investigations are much more numerous, and include work on gelatin, albumins, gums, rubbers, soap, serum, proteins, (especially H. Chick) and organic dyes. The principal conclusions may be summarised/

summarised as follows:-

(1) Emulsoids are unstable, giving an increase of viscosity with the passage of time.

(2) The viscosity of emulsoids is greatly altered by mechanical treatment. If they are shaken or passed through a capillary several times the viscosity decreases.

(3) As the concentration of emulsoid decreases the viscosity increases enormously and usually becomes quite unmeasurable at quite low concentrations, e.g. 2% to 3%.

(4) Added substances such as salts, acids or alkalies, alter the viscosity of many emulsoids to a very great extent. Most of the changes produced by added substances have hitherto defied analysis.

Regarding the mathematical theory of the viscosity relations in emulsoids and suspensoids little work has been done.

1906 Einstein deduces the formula

$\eta_1 = \eta_0 (1 + 2.2 f)$ where η_0 is viscosity of dispersion medium, η_1, viscosity of the suspension and f is the ratio of volume of solid phase to total volume.

1910 Bancelin finds an excellent agreement between theory and experimental results, using Einstein's formula, for gamboge particles.

1910 Hatschek deduces a formula very similar to that of Einstein, but containing the factor $4.5 f$ instead of $2.5 f$.

Smoluchowsky has criticised the derivation of these formulae very unfavourably. Hatschek has however obtained remarkable agreement between theory and experiment by the use of an instrument of the Couette type.

1911 Hatschek deduces the formula

$$\eta_1 = \eta_0 \left\{ \frac{\sqrt[3]{A}}{\sqrt[3]{A} - 1} \right\}$$

where η_1, is viscosity of system

η_0 viscosity of the continuous phase

A ratio of total volume to volume of disperse phase.

This equation fits the experimental results for glycogen, casein, and serum proteins quite well, but Smoluchowski considers it very unsatisfactory from a theoretical standpoint.

1916 Arrhenius shows by a large number of examples that the empirical formula $\log \eta = \Theta C$ expresses the viscosity of emulsoids with great accuracy. Θ is a constant and C is the "molecular concentration". This is found by making simplifying assumptions regarding the size of the molecules.

1921 Von Smoluchowski investigates the effect of electric charge on disperse phase. Such charge may affect the viscosity but experimentally the effect is negligible.

Considering the many similarities which exist between blood and simpler colloidal systems as regards their viscosity, it seems reasonable to approach the problem of the factors underlying viscosity of blood in a manner similar to that which would be used in investigating the viscosity of any other colloidal system.

As already observed, blood resembles a simple colloidal system in that its viscosity depends

(1) largely on the volume of the disperse phase, Burton-Opitz 1908-1906.

(2) On the presence or absence of substances which raise viscosity in the continuous phase. Dettermann 1908.

(3) In that Einstein's and Hatschek's equations are applicable. Murray-Lyon M.D. Thesis.

(4) In that viscosity falls with rise in temperature and in many other ways which will be shown in the experimental section below.

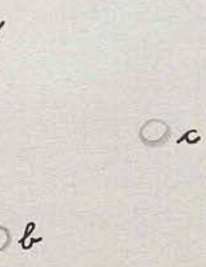
No satisfactory attempt has been made hitherto, to explain all the known variations of blood on physical principles and such indeed has probably been impossible until the present time, when perfected experimental methods and reliable data regarding simple systems render the attempt, at least a feasible one.

PART II.

THEORETICAL CONSIDERATION.

Before approaching the experimental part of the subject it is necessary that we should be quite clear regarding the factors which influence viscosity in systems containing a number of phases. We shall consider a number of simple cases.

CASE I. Consider three molecules a, b, c in a fluid. If a flow of such a nature as to move the molecule a, with respect to the molecules b, and c, is set up, work will require to be done in order to overcome the force which holds the molecule a, to the molecule b. The nature of this force is scarcely well known at present but is usually termed an "intermolecular attraction". When a is drawn out of the range of attraction of b it may then enter the range of attraction of c when it will fly towards c. The work done by overcoming the attraction between a and b manifests itself principally/



principally as heat, and since the amount of displacement of the molecules is a function of the force applied, the results in the form of flow, produced by the application of this force must also be related to the "intermolecular attractions" and so we have the relation of the applied force to the manifested results, giving rise to what we call a viscosity, or, to put it in simple terms, the viscosity is the resistance of the fluid to deformation or strain. It is related from a certain point of view to elasticity.

CASE II. Consider a fluid containing a rigid disperse phase, e.g. a kaolin suspension and compare this with the case of the fluid without the disperse phase. In this suspension, when we apply force to the entire system a distortion cannot occur in the rigid phase. A greater amount of distortion must therefore be produced in the continuous phase if we are to get the same result in terms of flow, hence either we must apply a greater force in order to get the same flow as in a pure liquid or else for the same force we get a lesser flow. That is, the presence of the disperse phase has the effect of increasing the viscosity of the liquid.

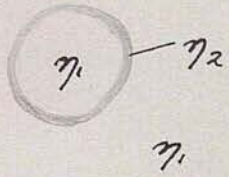
CASE III. If instead of the conditions in Case II we have a disperse phase which is partially rigid, its presence will obviously and for the same reasons, increase the viscosity of the whole system, for, if the applied force is able to produce only a very small deformation in the disperse phase, a great deformation must occur in the continuous phase which is equivalent to the appearance of a greater viscosity in the system.

An excellent example of this occurs in the case of a suspension of oil-droplets in water. The oil is much less deformable than is the water. The result is that if a flow is to be produced, the water must undergo a greater deformation than if the oil were not present. This gives a viscosity for the suspension, higher than that of water.

CASE IV. If the disperse phase is more distortable than the continuous phase, distortion will occur in it and therefore to a less extent than in the continuous phase. The net result will be that the mixture will show a lower viscosity than that of the continuous phase alone. We find an example of this in a suspension of ether droplets in water. The viscosity/

viscosity of the ether is small and therefore the viscosity of the whole system is small.

CASE V. Imagine now a system in which is a disperse phase of viscosity η_1, separated by thin envelopes of viscosity η_2 and dispersed through a continuous phase, again of viscosity η_1. Plainly from the foregoing any increase or decrease in the viscosity of the whole system compared with the viscosity of the continuous phase will be a measure of the viscosity of the envelopes. If the envelopes are not fluid but solid, their viscosity will not be measured but the corresponding property applicable to solids, i.e. deformability will be measured instead, in terms of a viscosity. A good case of this rare system is the case of air-bubble in air, each bubble surrounded by a thin layer of soap-solution η_2 which differs from the air which is both inside and outside the investing envelope. Here the viscosity of the whole system is greater than that of air and greater/



greater by an amount which corresponds to the deformability of the investing membrane.

CASE VI. This is the case of red blood-cells in saline suspension and is very like the preceding case, except that now the fluids on the inside and the outside of the cell envelope are not of the same deformability. Now two factors increase the viscosity of this system above that of saline.

(1) The deformability η_1 of the fluid inside the cells, which is greater than that of the saline η_2.

(2) The resistance to deformation of the envelopes, which measured as a viscosity η_3 is even greater than η_1.

It therefore follows that the viscosity of a suspension of red cells is determined by:-

(1) the deformability of the external medium, saline or serum.

(2) the deformability of the corpuscular contents.

(3) the resistance to deformation on the part of the envelope.

We may now proceed to tabulate the factors which we would expect on theoretical grounds to play a part in the production of a viscosity in a complex system such as a suspension of red-cells in saline.

(1) The viscosity of the continuous phase, e.g. plasma, serum or saline.
This, it is to be noted, constitutes a system in itself, for it too has a continuous phase of water and salts and a disperse phase of albumins, globulins, fibrinogen, and other colloidal substances.

(2) The viscosity of the corpuscular contents which again constitute a complex system with haemoglobin as a disperse phase.

(3) The resistance to deformability of the cell envelopes.

(4) The volume of the corpuscular phase.
It will be quite obvious from the consideration of all the cases which we have considered that this is a most important factor, for, as the viscosity is dependent on deformability of the respective phases, an increase in the volume of any one phase will obviously produce a greater viscosity in the whole system.

(5) Since it is the resistance to deformation which is responsible for the appearance of a viscosity, it will be clear that any factor which makes the corpuscular phase more readily deformable will have an effect on the viscosity of the whole system.

One/

One of these factors is, of course, that mentioned under (3) but another equally important is the shape of the envelopes and the enclosed content. This may be shown in the following way. When a flow is set up in a suspension of cells, the cells orientate themselves broadside on to the flow. They do this because this is the position of greatest stability and it is well known on hydro-dynamical principles that it is also the position of greatest resistance. An applied force of such a nature as to cause a flow will therefore be resisted by this position of the cells to a greater extent than by the cells in any other position, and this will have its effect on the viscosity of the entire system. This viscosity will be higher than that which would result were the cells arranged haphazard or if the cells were in a spherical form.

(6) A special case of the preceding (5) is where the cells are not single but in aggregates or rouleaux. Here again the arrangement of the cells in the stream will affect the viscosity of the system although the effect is a very complex one. It will be alluded to again in the experimental section. /

section. In general it must lessen the viscosity, if for no other reason than that if the cells are in large aggregates they must at least be clear of the periphery of the flowing stream.

(7) Von Smoluchowski has shown that there is in theory at least an effect on viscosity due to the electric charge. Such charges constitute additional forces, to overcome which additional work must be done. He has however shown that the effect of these charges, unless very great, is negligible compared to the other forces involved.

P A R T III.

PART III.

METHODS.

(1) Ostwald Method.

(2) Couette Method.

(3) Dettermann's Method.

(4) Hurtle's Method.

(5) Hess's Method.

In considering method we shall deal only with those methods which are applicable to blood, serum and suspensions of red-cells and only with those methods which have some claims to be considered accurate.

(1) The Ostwald Method. This consists of a U tube, one of the limbs being narrowed down to a fine capillary from .5 to 5 mm. in diameter. At the upper end of the capillary is a bulb of small capacity, about .5 to 1 cc. capacity. Above the bulb the capillary is continued and opens to the air. A few mm. above and below the bulb, the capillary is etched with a mark. The other limb is wide and has at its lower end near the U a bulb of about 3 to 5 cc. capacity. It is convenient to attach a tube with a mouthpiece to the wide limb. The whole instrument is attached to a metal support, kept/

kept perfectly vertical by means of a hanging weight and is immersed almost completely in a thermostat at any required temperature. The instrument is first calibrated with pure distilled water. The water is introduced into the bulb on the wide limb and by blowing through the mouthpiece is forced up the narrow limb, filling its bulb and into the capillary above the bulb. The purposee of this is to wet the apparatus thoroughly. The pressure may now be released, when the fluid falls in the capillary limb. When it has attained the temperature of the thermostat it is once more forced up to above the upper mark in the capillary limb. The pressure is now released and the time taken for the upper meniscus of the column to pass between the two marks is noted. Six determinations should be made. These should not differ by more than + or - 1%. E.g. we found with one of our instruments, the following figures for pure distilled water, were obtained:-

45.2 secs.
45.2 secs.
45.0 secs.
45.2 secs.
45.4 secs.
45.2 secs.
Mean 45.2 secs.

Here/

Here a very important point should be observed. If the viscosity of the fluids to be examined is low, an instrument giving as great an outflow time as possible should be used, e.g. one with a capillary bore of .5 mm. If on the other hand we are to work with whole blood where the viscosity is high , it is advisable to use an instrument giving a short outflow time for water, e.g. 20 secs.

If we use, for blood, an instrument with an outflow time of more than 30 secs. the outflow time for blood becomes very long. The question now arises as to whether the viscosity of a fluid as compared with that of water differs according to the bore of the capillary used.

The following experiments show that it does not.

Instrument I.

Bore about .5 mm.

Outflow time for water 45 secs.

Outflow time for blood 243 secs.

Relative viscosity $= \frac{243}{45}$

$= 5.166$

$\eta = .0465$

Instrument II.

Bore about 1 mm.

Outflow time for water 27 secs.

Outflow/

Outflow time for blood 138 secs.

Relative viscosity $\frac{138}{27}$

$= 5.111$

$\eta = .0460$

Instrument III.

Bore about 3 mm.

Outflow time for water 12 secs.

Outflow time for blood 61 secs.

Relative viscosity $= \frac{61}{12}$

$= 5.08$

$\eta = .0457$

Instrument IV.

Bore about 5 mm.

Outflow time for water 7 secs.

Outflow time for blood 34 secs.

Relative viscosity $\frac{34}{7}$

$= 4.857$

$\eta = .0437$

The blood used in these experiments was defibrinated ox blood. It will be seen that the viscosity as measured by instruments showing a ten-fold variation of capillary bore is constant at the figure of five.

The absolute viscosity is found by multiplying the absolute viscosity for water at 25°C which is 0.009 by this/

this value 5.

The Ostwald Method possesses several very marked advantages and disadvantages.

Advantages.

(1) It is, of all viscosimeters except possibly the Couette type, as modified by de Nouy , the most accurate. The errors arising in its use should not amount to more than + or - 1%.

(2) The calibration is very simple. All that is required is to take the outflow time for distilled water. This calibration being so simple can be repeated from time to time as a check.

(3) The instrument being inexpensive, it is possible to possess a wide range of varying bore, for use with fluids of widely varying viscosity.

(4) The results are characterised by a remarkable constancy owing to the fact that the driving force is gravity acting in the vertical plane.

(5) The method requires only small quantities of blood, .1 cc. to 2 cc. being sufficient.

Disadvantages.

(1) It is necessary to use blood to which an anticoagulant has been added. This is a disadvantage which cannot be overcome, for, if anticoagulant is absent, the blood will clot in the capillary.

Burton-Opitz and others have objected to the use of/

of anticoagulants on the ground that they tend to lower viscosity. This particular disadvantage of the Ostwald method, is of importance only, in the investigation of changes which arise in vivo.

When purely in vitro investigations are carried out, the objection is much less serious. The difficulty cannot be overcome by the use of defibrinated blood, for the viscosity of this, as Burton-Opitz has shown, is much less than that of fresh blood. This is only what might be expected, for defibrination removes fibrinogen, an important protein component of the blood.

(2) The method is liable to error because of lack of attention to certain small details. The most important of these are:-

(a) The failure to keep the instrument perfectly vertical.

(b) The presence of grease in the capillary.

(c) The presence of froth at either meniscus.

Grease is best removed by washing with alcoholic KOH , then with HNO_3 and finally by blowing steam through the instrument. The apparatus should then be dried in a hot-air oven in order to prevent small quantities of water, from diluting the material to be measured.

(3) The necessity of blowing the fluid up the capillary involves the creation of a new surface in the fluid. Gokun and Biltz have shown that in the case of emulsoids, even the passage through a capillary gives/

gives rise to changes of viscosity. This they attribute to the fluids, possessing a certain "structure". The differences however are not very marked.

Various modifications of the Ostwald apparatus have been devised. For the most part they are distinguished by alterations in unimportant detail.

The use of a small viscosimeter, based on the Ostwald principle, has been suggested for clinical work. It consists of a small funnel from which there leads a capillary into a U tube. Blood is dropped from the ear or finger into the funnel and the time taken for it to flow through the capillary, measured. The instrument is calibrated with water, but the calibration is fallacious, because it is much easier to fill the funnel with water than with blood.

2. Dettermann's Method. The Dettermann instrument introduced in 1908 consists of two bulbs separated by a long straight capillary. The instrument is enclosed in a water-jacket made of glass, from the ends of which, tubes leading into the bulbs, project.

The instrument complete with water-jacket, rotates about its middle point in a vertical plane. The capillary bears two marks, equidistant from the ends of the capillary.

To use the instrument the water-jacket is filled with/

with water at the desired temperature. Water is then introduced into the lower bulb, the instrument rapidly inverted and the time taken for the meniscus to pass between the marks measured. This is the outflow time.

To measure the viscosity of blood or any other fluid, the fluid or blood is placed within the bulb and the outflow time measured as in the case of water. The ratio of the outflow time for water gives the relative viscosity as in the case of the Ostwald instrument. The apparatus must be used with defibrinated blood or blood containing an anticoagulant. Murray-Lyon in his researches employed this apparatus and used blood which contained Hirudin. He states that this substance does not alter the viscosity but in this statement is not in agreement with Burton-Opitz.

From its construction it will be clear that the Dettermann instrument possesses more disadvantages than does the Ostwald and at the same time has few advantages. The small water-jacket is unsatisfactory, the rotation of the instrument causes forces more complex than in the case of the Ostwald, to act on the column of blood, and, most important of all, the instrument is very difficult to clean. There is no compensating advantage for preferring this apparatus to the much simpler Ostwald viscosimeter.

(3)/

(3) The Couette Viscosimeter. This method is based on the investigations of Saint-Venant, Stokes, Margoules and Couette. In principle it depends on the fact that two concentric cylinders separated by a layer of blood have motion imparted from one to the other according to the greater or lesser viscosity of the blood separating them.

The apparatus as improved by Hatschek and by de Noüy consists of a small metal cylinder, rotating about a vertical axis. The speed of rotation is constant and the drive is supplied by a constant speed electric motor. Within this cylinder which may be about a centimetre in radius, hangs a second cylinder with a radius of about 1.6 mm. This second cylinder is usually solid and tapered to a conical point at its lower end. It hangs on a fine suspension which may be made of phosphor-bronze or of steel according to the strength required. On the suspension is mounted a mirror and at the upper end an adjusting screw. The mirror reflects a beam of light onto a scale divided into degrees. The whole apparatus is contained either within an air thermostat or within an oil-bath at 37°C. The blood is introduced into the space between the two concentric cylinders until it covers the inner cylinder to a certain marked level. The outer cylinder is then set rotating and the movement/

movement of the inner cylinder measured in degrees of arc. The inner cylinder tends to rotate with the outer cylinder owing to the fact that at neither boundary does the fluid slip, the boundaries being thus fixed with respect to the portions of fluid very near to them. Any deformation which occurs must occur in the fluid itself. This deformation is a viscosity and the greater the deformation the less will be the movement of the two clyinders with respect to one another.

In the case of blood the suspension must be a heavy one, preferably of flat steel ribbon. The blood is run from the vessel of the animal through a paraffined canula into the cup which may contain hirudin or oxalate. Alternatively defibrinated blood may be used.

The instrument is calibrated with distilled water, the deflection corresponding to the fluid being found at a known speed of rotation. The viscosity of the blood is then,

$$\frac{\text{scale reading for blood}}{\text{scale reading for water}} = \frac{\eta_1}{\eta_0}$$

η_1 = viscosity of blood in units relative to η_0 which is viscosity of water.

The advantages of the Couette viscosimeter are several. Measurements can be taken without continual creation of new surface in the blood and thus one of the/

the errors attached to the use of the Ostwald instrument is done away with. The apparatus, once calibrated, is very easy to work and has the distinct advantage, that by varying the speed of motion of the external cylinder the same instrument can be used for fluids of widely varying viscosity. The apparatus on the other hand is associated with certain disadvantages.

The principal of these, is that even when defibrinated blood is used there is a tendency for the blood to separate into concentric layers. Although de Noüy and others regard the method as very accurate for the determination of the viscosity of certain colloidal solutions they do not recommend its use in its present form at least, in the case of blood.

(4) Hurtle's Method. is the method by which Burton-Opitz has carried out all his investigations. It is the only method in which an attempt is made to measure the viscosity of the blood in the living animal.

A canula is inserted into one carotid artery and this is attached to a capillary tube about 10 to 12 cm. long and of known diameter. This tube delivers the blood which flows through it into a tilting recorder which records the delivery of .5 cc. of blood at a time. Burton-Opitz uses the tilting recorder to work an electric signal writing on a drum. From the length

and/

and diameter of the capillary and from the outflow time the viscosity of the blood is calculated by means of Poiseuille's law. In order that the pressire at which the fluid is delivered shall be known, a canula attached to a manometer is inserted into the other carotid artery.

From its very nature this method can dispense with the use of all anticoagulants. The temperature at which the viscosity is measured is 37°C. as this is the temperature of the blood as it leaves the animals vessels. As a precaution against change of temperature Burton-Opitz surrounds the canula by a small water-jacket. The variations of pressure due to the beat of the heart are so small as to exert no effect and providing that clotting does not occur in the capillary or the canula, readings can be taken again and again.

The one disadvantage of the method is, that owing to the haemorrhage which must accompany its use, the blood-pressure is continually falling. It is difficult to use except in animals under anaesthesia and is said to be very liable to errors due to the formation of small clots which are only with difficulty detected. The apparatus is obviously quite useless for in vitro experiments and thus cannot be used with any degree of accuracy for experiments which are chiefly concerned with the physical factors determining viscosity.

(5)/

(5) Hess's Method. This method is a modification of the methods whereby viscosity is measured in terms of the time taken for a fluid to flow through a capillary tube. The apparatus consists of two capillary tubes of equal bore lying parallel, the one dipping into a cup of water and the second dipping into a cup containing the blood. By means of a rubber bulb communicating with both tubes, the blood and water are sucked up simultaneously, the difference of rate being observed on a scale attached to the tubes.

This method has no particular advantage. Anti-coagulants are necessary and the instrument has the actual disadvantage that it is very clumsy.

In view of the facts that:-

(1) The Couette instrument has not been much used for the viscosity of blood.

(2) That Hurtle's method is suited to experimental animals only.

(3) That the methods of Hess and Dettermann possess no advantage over Ostwald's method,

we have selected this method for use in our investigations.

Of all methods it is the simplest, the most direct and the least liable to error in the case of blood and suspensions of blood-cells.

P A R T IV.

PART IV.

EXPERIMENTAL.

Section I.

As indicated in Part II this section deals with the first of the factors which influence the viscosity of blood, viz. the viscosity of the continuous phase which is plasma in the case of circulating blood, serum in the case of defibrinated blood and usually, saline in the case of a cell suspension.

The observations on the viscosity of serum and plasma are not very numerous, partly because the subject is of comparatively little interest when separated from the greater subject of the viscosity of the blood. The principal authorities are Josue and Pasteurier who give observations on the viscosity of the serum and plasma of many animals and who find that in the mammalia at least, the differences in the viscosity of serum and plasma between the members of different species, are comparatively small.

We have investigated the following points.

(A) Viscosity of normal oxalated plasma.

Oxalated plasma is as nearly representative of the plasma in the blood-stream as it is possible to obtain/

obtain provided that the amount of added oxalate is small.

Determinations of the viscosity of such plasma from the blood of various normal animals gave the following results:-

Man.	(1)	.0155
	(2)	.0158
	(3)	.0150
	(4)	.0162
	(5)	.0157
Rabbit.	(1)	.0102
	(2)	.0108
	(3)	.0103
	(4)	.0107
	(5)	.0115
Ox.	(1)	.0164
	(2)	.0172
	(3)	.0168
	(4)	.0173
	(5)	.0163
Horse.	(1)	.0188
	(2)	.0194
	(3)	.0182
Guinea-pig.	(1)	.0114
	(2)	.0122
	(3)	.0118
	(4)	.0116
	(5)	.0125

Goat./

Goat.	(1)	.0174
	(2)	.0169
Sheep.	(1)	.0184
	(2)	.0192
	(3)	.0186
	(4)	.0183
Rat.	(1)	.0114
	(2)	.0106
	(3)	.0109
Pig.	(1)	.0154
	(2)	.0161
	(3)	.0158
	(4)	.0156

The specimens examined, five in number, in most cases were obtained from animals used in the Department of Bacteriology. All the animals, were, as far as could be ascertained, normal. Small quantities of blood (about 5 cc.) were withdrawn into small flasks containing 5 mgm. of potassium oxalate, in a state of fine powder and the plasma separated off by centrifugalisation for 10 minutes at 2000 revs. per min. The measurements were all carried out at 25°C., the viscosity for water at this temperature being 0.0009.

It will be seen from the results that the differences between the viscosities of the plasma of different mammals are not very great. In no case is the viscosity/

viscosity of the plasma greater than 1 - 7 times that of water. Since viscosity of blood is as a rule more than 4 times that of water the viscosity of the plasma is obvicusly a matter of minor importance.

It will be understood that the figures given above are values of η in absolute units.

(B) The viscosity of serum.

Proceeding in a similar manner to the investigation of the viscosity of plasma we have measured the viscosity of the serum of the same series of animals.

The blood was drawn into a test-tube and allowed to clot. The clot was then "wrung" and allowed to stand over-night in the ice-chest at 0°C. The supernatant serum was then drawn off and its viscosity determined at 25°C.

Man	(1)	.0139
	(2)	.0145
	(3)	.0137
	(4)	.0144
	(5)	.0141
Rabbit	(1)	.0093
	(2)	.0096
	(3)	.0092
	(4)	.0095
	(5)	.0099

Ox/

Ox.	(1)	.0148
	(2)	.0154
	(3)	.0152
	(4)	.0157
	(5)	.0147
Horse.	(1)	.0172
	(2)	.0173
	(3)	.0166
Guinea-Pig.	(1)	.0102
	(2)	.0108
	(3)	.0108
	(4)	.0104
	(5)	.0111
Goat.	(1)	.0154
	(2)	.0150
Sheep.	(1)	.0164
	(2)	.0174
	(3)	.0168
	(4)	.0164
Rat.	(1)	.0102
	(2)	.0098
	(3)	.0100
Pig.	(1)	.0138
	(2)	.0147
	(3)	.0144
	(4)	.0141

In some cases the blood was drawn into a small flask containing glass-beads. The flask was rapidly agitated for a period of 15 minutes and allowed to stand overnight. The serum was then separated off by centrifugalisation for 10 mins. at 200 revs. per min.

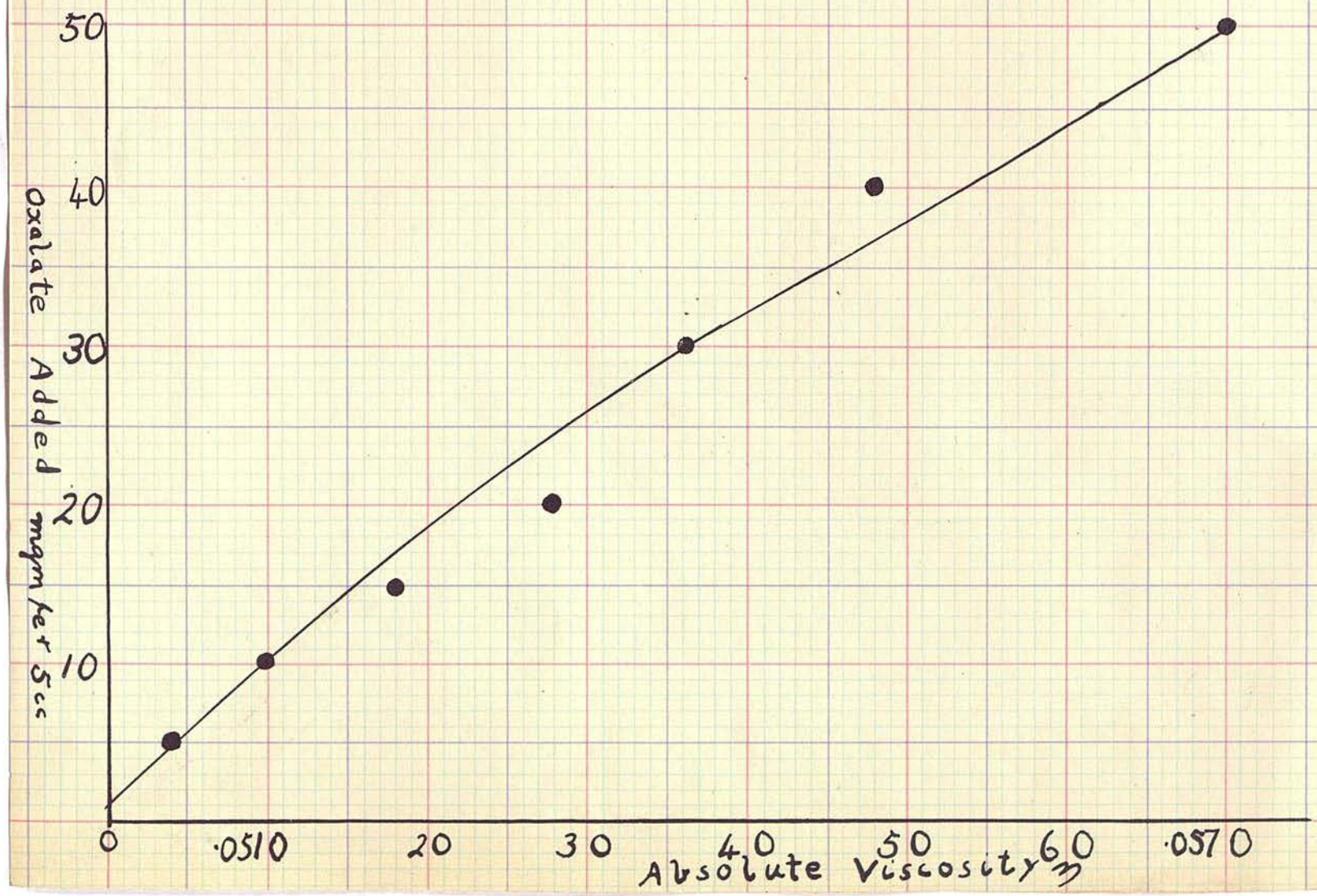
In all the cases the viscosity of the serum was noted to be about 10% less than that of the plasma. This difference is obvicusly greater than any reasonable allowance for experimental error, which is normally about the range of 2%. The simplest explanation for this phenomenon is that the fibrinogen, which being a protein, contributes to the viscosity of the plasma, is no longer present in the serum and therefore the viscosity of the serum is less, owing to the fact that its disperse phase possesses a smaller volume than that of the plasma.

(C) <u>The effect of various anticoagulants.</u>

(1) Oxalate. It is onvicusly of great importance that we should know the effects of small and large amounts of oxalate on the viscosity of the plasma. Accordingly we added different amounts of oxalate ranging from such an amount as was just sufficient to prevent coagulation, to that amount which was sufficient to produce actual lysis of the cells themselves.

As/

GRAPH of EFFECT of ADDED OXALATE on THE ABSOLUTE VISCOSITY of RABBIT BLOOD



As the following figures show, the amount of oxalate used has an important bearing on the results obtained. The amounts of oxalate shown below are the quantities added to 5 cc. of blood and the viscosities are the viscosities of the separated plasma.

In all the experiments quoted below, the blood used was that of the rabbit.

Oxalate added.	η
5 mgms.	.0504
10 mgms.	.0510
15 mgms.	.0518
20 mgms.	.0528
30 mgms.	.0536
40 mgms.	.0548
50 mgms.	.0570

From the above results it will be noted that quantities from 5 mgms. to 15 mgms. of oxalate exert little or no effect on the viscosity of the plasma. Larger quantities we find increase the viscosity to a considerable extent. The very marked increase in viscosity which takes place when 50 mgms. of oxalate are added to 5 cc. of blood is apparently due to the fact that lysis occurs to a considerable extent as evidenced by the free haemoglobin in the plasma. This haemoglobin being an added protein increases the volume of the dispersed phase and thus increases

the/

the viscosity. This result is not entirely unexpected in view of the fact that Burton-Opitz finds in his experiments that the addition of any quantity of oxalate exerts an effect on the viscosity of the blood.

(2) Sodium Citrate. In this case the blood was run into a flask containing finely powdered sodium citrate. The quantity was 15 mgms. to 10 cc. of rabbits' blood. Smaller quantities than this, we found, did not give satisfactory results, owing to the premature clotting of the blood.

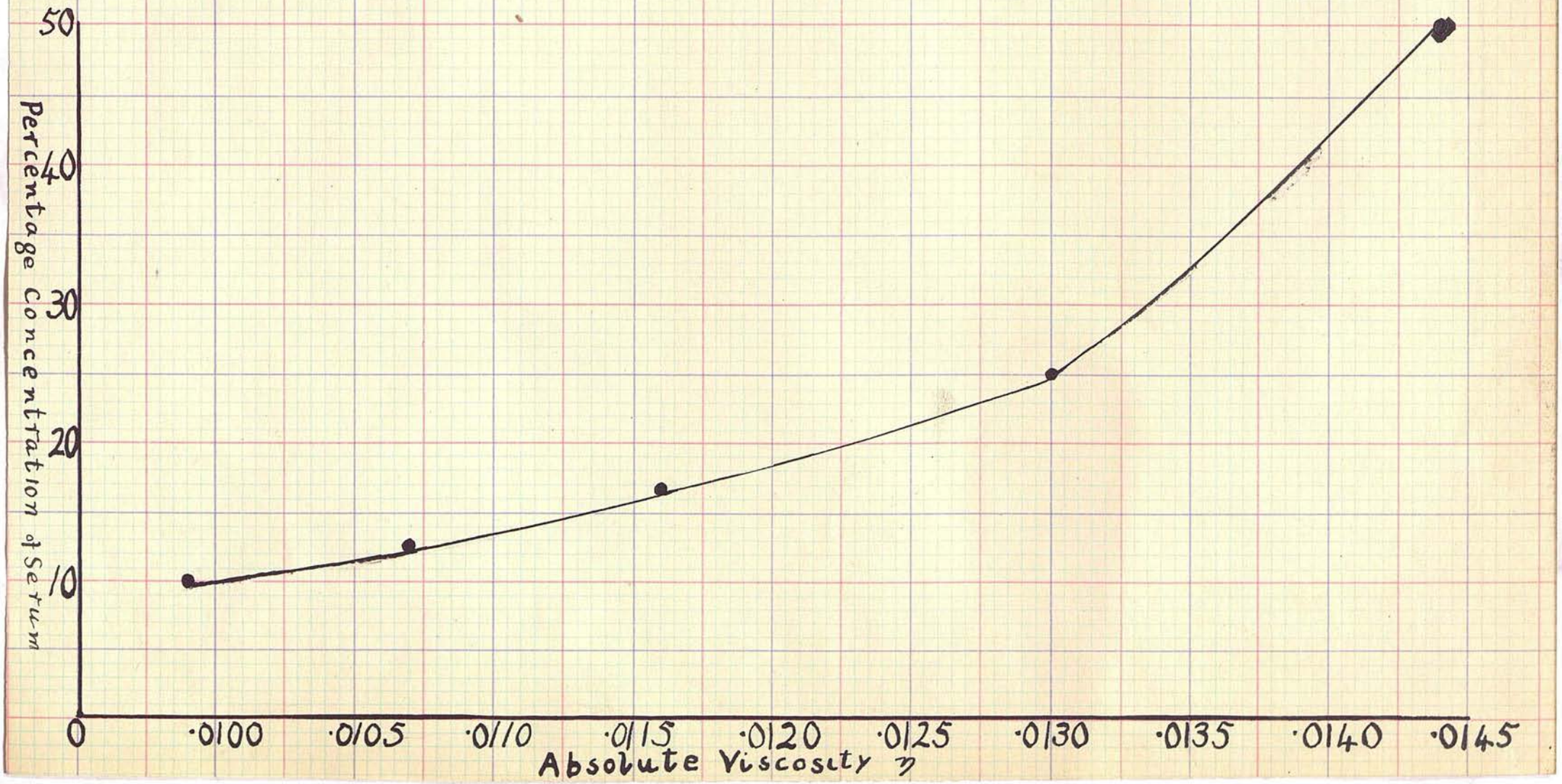
For rabbit's blood we found the value of η to vary between .0512 and .0523.

This is no greater than the results obtained, using potassium oxalate.

(D) The effect of various dilutions on the viscosity of the plasma.

The investigation of this problem is very similar to the investigation of the problem of the effect of dilution on the viscosity of any emulsoid, except that in the case of water as a diluent the globulins are immediately precipitated and the viscosity falls accordingly, for the previously dispersed emulsoid phase now becomes a suspensoid phase and, as we have previously shown, a suspensoid phase exercises little effect/

GRAPH SHOWING EFFECT of DILUTION on the VISCOSITY of OX SERUM.



effect on the viscosity of the system. The diluent must therefore be a sodium chloride solution in which the globulins are soluble. .85% sodium chloride is satisfactory.

The effect of dilutions from 1 in 2 to 1 in 10, on ox serum are shown in the following table.

Serum.	η
1 in 2	.0144
1 in 4	.0130
1 in 6	.0116
1 in 8	.0107
1 in 10	.0099
.85% saline	.0095

As the dilution progresses we note that the viscosity of the serum steadily approaches that of the diluting fluid. Again the volume of the disperse phase is reduced with a corresponding fall in the viscosity.

It will be noted in the graph, which is attached, that, in the higher dilutions in the region of 1 in 10 to 1 in 5, the viscosity changes to a very small extent, but, as the range of concentration is increased, the viscosity changes very rapidly.

The dilution of the serum is of considerable theoretical importance because, after severe haemorrhage, the volume of the blood is made up by a passage of fluid from/

from the tissues into the blood-stream, which of course dilutes the plasma.

It will be further seen from the graph that slight changes in the region of fully concentrated serum produces a relatively great change in the viscosity and that therefore even slight degrees of dilution may be quite important.

(E) The effect of heat on serum.

Ox and rabbit serum were heated to 56°C. for ½ hour in a constant temperature bath. This temperature was chosen (a) because it is the temperature for inactivation of complement and (b) because it is the temperature at which the serum produces the effect of marked rouleaux formation. The temperature is of course below that which is required for precipitation of any of the proteins. Although a considerable number of experiments were done in the case of each animal, the results obtained were very irregular. In a number of cases the viscosity was found to be slightly higher, in a number slightly less, no great differences were however observed and no uniformity attained.

SECTION II./

SECTION II. This section deals with the second factor which enters into the question of the viscosity of the whole blood, viz. the viscosity of the corpuscular contents which are fluid in nature.

To obtain this viscosity we must first obtain the corpuscular contents. This was obtained by the following method. A quantity of ox-cells was washed very thoroughly and after the last washing with saline the cells were centrifugalised for ½ hr. at 3,000 revolutions per minute. By this means all the saline was obtained supernatant to the cells and was easily removed. The closely packed cells were then frozen rapidly in the refrigerator.

They were then thawed rapidly by plunging the tube and contents into warm water. This process was repeated three times so that the final product consisted of cell contents and cell envelopes.

The mixture was again centrifugalised very thoroughly and the supernatant fluid used for the experiments. The viscosity was measured as before, at 25°C. and the following results obtained.

Experiment I. .040

Experiment II. .043

Experiment III. .037

These/

These experiments agree in showing that the viscosity of the corpuscular contents is about 4.5 times that of water. This result is very high and it must be admitted that the experiments are not very satisfactory in that the yield of corpuscular contents is very small. There is, however, no reason to suspect that the corpuscular contents are particularly altered by the process of manufacture.

Since the haemoglobin is in about 30% concentration, within the cell, this high result is not so very remarkable as it at first appears.

In order to see whether a 30% solution of haemoglobin in saline possessed comparable viscosity, we attempted to make such a solution with a specimen of crystalline Haemoglobin. The haemoglobin however was not soluble to that extent. A 15% solution was found to give a viscosity 2.7 times that of water.

Since the requirements of the corpuscular contents is so high it is quite plain that the corpuscules, even if they were not surrounded by a membrane, would show a large viscosity effect, but, as they are surrounded by a membrane and therefore the haemoglobin droplets are less deformable, and, as the number of such droplets per unit volume of blood is very great, the viscosity of the blood must depend to a large extent on the viscosity of the corpuscular contents.

SECTION III. This section deals with the third factor which enters into the viscosity of the blood, viz. resistance to deformation, of the cell envelopes.

It is a matter of great difficulty, to measure this factor, for, in order to do so it is necessary to obtain the cell envelopes in suspension in saline. By measuring the viscosity of the saline and then by measuring the resistance of the suspension, the difference in the two results gives the resistance to deformation of the suspension.

The method adopted was as follows. The cells from 10 cc. of defibrinated ox blood were thoroughly washed several times with saline and the washed cells were then haemolysed with three times their volume of water. The cell envelopes which are not destroyed by this process were then centrifugalised down on a high-speed centrifuge and washed twice with saline. They were then re-suspended in 10 cc. of saline, an amount equivalent to the amount of blood from which they were originally obtained.

The viscosity of this suspension was then compared at 25°C. with that of saline.

Saline .0095

Suspension .0135

Viscosity/

Viscosity of the suspension is thus 1.5 times that of saline, which is very nearly the same as that water, viz. .0098 this extra viscosity, must be due to the resistance to deformation of the cell envelopes.

It is obvious that this may not represent the contribution to the viscosity of the blood, by the envelopes of the intact cells, for the following reasons.

(1) There is no guarantee that the treatment to which the cells were subjected has not altered the resistance to deformation of the envelopes.

(2) That the form of the envelope in the saline suspension must necessarily be different from the shape of the envelope in blood.

The contribution of the resistance to deformation of the envelope to the viscosity of the whole blood must of reason be rather small, although it must certainly be taken into account.

An analogous case to that of the above might be mentioned. It is well known that a suspension of air-bubbles in air, is in such a state that individual soap membranes enclosing the air are easily deformable. Now, suppose that the interior of the soap membrane is filled with a viscous substance of viscosity much greater than air, then the deformability of the membranes will be much less. The reason for this is that any deformation of the membrane necessarily produces a deformation/

deformation of the internally contained substance and this, resisting deformation to a greater extent than does the membrane itself, will play the more important part in determining what deformation will actually result from the application of a given force.

In precisely the same way the contents of the red blood cell envelopes resist deformation to a much greater extent than do the envelopes themselves. Accordingly the effect on the viscosity of the whole system is the chief factor involved.

Now, if it were possible to remove the envelopes without destroying the continuity of the contents we would obtain a state analogous to that of oil-droplets in water, where, as we have shown above in the part devoted to theoretical considerations, the viscosity of the system is increased by the fact that the oil possesses a high viscosity.

While in practice it is not possible to remove the envelopes, in theory it is perfectly allowable, for the experiment quoted above shows that the contribution of the envelope to the total viscosity of the system is quite small.

It is unfortunately impossible to alter the resistance to deformation of the envelopes in any certain manner.

SECTION IV. This section deals with the volume of the corpuscular phase and its influence on the viscosity of the blood or suspension.

The volume of the corpuscular phase is determined by two factors:-

(1) Number of cells present

(2) Size of cells.

We shall first consider the influence of number of cells on the viscosity of the suspension.

(1) Number of cells present.

It is obviously of great importance that the shape of the cells should be known for we have already pointed out, in the theoretical considerations that shape is a factor in determining the viscosity and this factor must therefore be excluded in considering the influence of the number of cells on the viscosity of the suspension.

Another factor which must be considered is that of serum. It is necessary in determining the effect of the number of cells on the viscosity of the suspension, that the cells should be suspended in normal saline and not in serum, for, if serum were used, the cells would tend to go into rouleaux formation and moreover the additional factor of the viscosity of the serum would be introduced.

In/

In saline, the cells will not form any aggregates but will take up a definite form described by Gough. This form is a convenient one, for it makes the cell comparable to a droplet of oil.

Great care must be taken to determine the number of cells in the fluid. This is done by counting the cells present in the saline suspension by means of a Thöma Haemacytometer. The suspensions are then reduced to a standard cell content.

This content we have decided to make 2×10^{14}.

The following viscosity determinations were then made, for the blood of the horse, rat, pig, rabbit, guinea-pig, ox, and man.

In order to show the accuracy of the determinations the results are given in full.

	Outflow time.	Rel. Viscosity.	Abs. Viscosity.
Horse	18.6		
	18.6		
	18.6		
	18.6	1.087	.0098
	18.6		
	18.5		
	18.6		
Rat	18.6		
	18.4	1.087	.0098
	18.2		
	18.4		

Pig. /

	Outflow time.	Rel. Viscosity.	Abs. Viscosity.
Pig	18.9		
	18.9	1.099	.0099
	18.8		
Rabbit	18.0		
	18.0	1.048	.0094
	18.0		
	18.1		
G.-Pig	18.6		
	18.6	1.087	.0098
	18.6		
Ox	18.1		
	18.1	1.053	.0095
	18.1		
Man	18.0		
	18.0	1.048	.0094
	18.0		

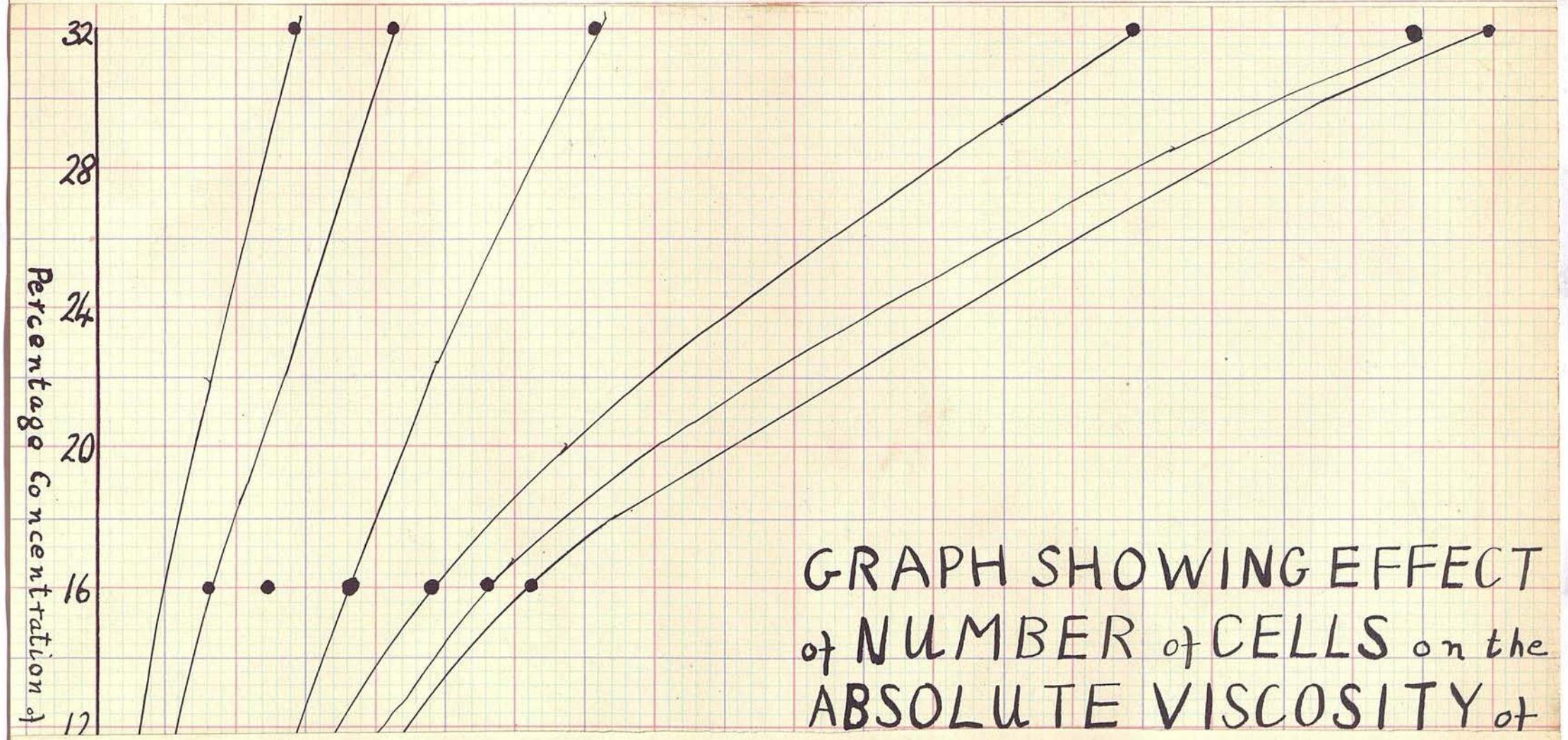
From the above results we see that differences are so small and inaccurate that it is of little use investigating this strength of suspension which is that which is used generally in serological and haematological work. Accordingly to obtain more conclusive results we have increased the strength of the suspension with the following results.

The /

The results are given as the average of three outflow times with each strength of suspension.

		Rel. Viscosity.	Abs. Viscosity.
Sheep	32%	2.6	.0234
	16%	1.58	.0142
	8%	1.18	.0106
	4%	1.09	.0098
Ox	32%	4.7	.0423
	16%	1.89	.0170
	8%	1.25	.0112
	4%	1.09	.0088
G.-Pig	32%	5.80	.0522
	16%	2.12	.0191
	8%	1.33	.0119
	4%	1.18	.0106
Horse	52%	1.75	.0157
	16%	1.23	.0111
	8%	1.09	.0098
	4%	1.06	.0096
Man	32%	6.10	.0549
	16%	2.29	.0206
	8%	1.47	.0132
	4%	1.24	.0111
Goat	32%	1.34	.0121
	16%	1.111	.0099
	8%	1.07	.0096
	4%	1.05	.0955

By/



GRAPH SHOWING EFFECT of NUMBER of CELLS on the ABSOLUTE VISCOSITY of

By 4% suspension is meant a suspension containing 2×10^{14} cells per cubic centimetre. This is the result of making up the washed blood cells from 4 cc. of blood to 100 cc. with normal saline. Hence the expression in terms of percentage.

The curves are shown in the attached graphs.

From the results we draw the following conclusions.

(1) The viscosity of a suspension increases with the cell content. This is what we expect from theoretical considerations, for we have a greater volume of rigid or semi-rigid phase in the system.

(2) The viscosity increases comparatively slowly with the increase in cell-content until about 20% of the cells are present. Thereafter the increase in viscosity occurs very rapidly. This fact is important because, as in the case of the circulating blood, where we have a 40% cell content, a small change in cell content will produce a relatively great change in viscosity. If however the circulating blood were very dilute, say of 4% concentration, a comparatively large change in cell-content would produce a small change in viscosity.

We/

We have attempted to show that Hatscheck's formula

$$\eta_1 = \eta_0 \left\{ \frac{\sqrt[3]{A}}{\sqrt[3]{A} - 1} \right\}$$

Is applicable to the above results for this formula is stated to give the relation between viscosity and the quantity of solid phase.

We have found, however, that the formula does not satisfactorily fit the results, the difficulty being that the increase of viscosity is not sufficiently great in the experimental results when the cell concentration is low, and further, that it is too great when the cell concentration is high. The reason for this we were unable to explain.

It is possible, however, that it is connected with the fact that the cell itself is considerably deformable.

(2) The size of cells.

In order to investigate the effect of the size of the cells on the viscosity it is firstly necessary to make the number of cells constant.

The numbers were made constant and equal to 8×10^{14} cells per cc. This number has been decided upon, for we have already shown that the viscosity of cells of number 2×10^{14} is very near that of water and therefore/

therefore not suitable for measurement. We have also shown that suspensions containing 16 x 10^{14} cells per cc. have a very high viscosity and are therefore also, not suitable.

Animal	Size.	Vol.	Rel. Visc.	Abs. Visc.
Sheep	4.8 μ	20 μ^3	1.58	.0142
Horse	5.5 μ	26 μ^3	1.23	.0111
Ox	6.0 μ	33 μ^3	1.89	.0170
G.-Pig	7.2 μ	58 μ^3	2.12	.0191
Man	7.8 μ	78 μ^3	2.29	.0206

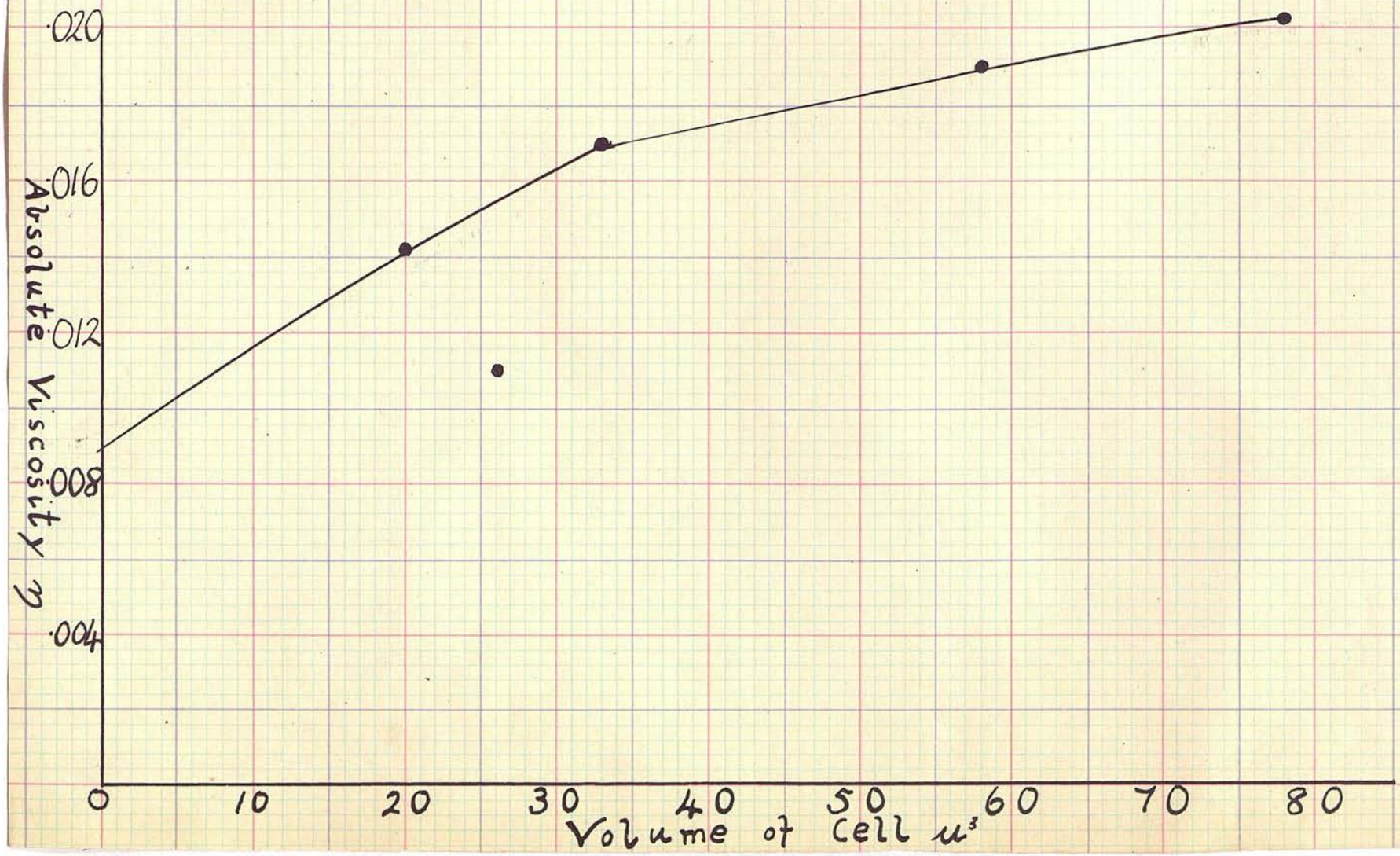
Two points must be noted:-

(1) The diameters and volumes quoted above were obtained from "The Erythrocyte and action of simple Haemolysins" and are for dried cells. These figures do not apply to cells in suspensions in the fresh state but are proportional to the diameters and volumes of the fresh cells.

(2) The diameters are for cells in the disc-like state, whereas the cells in these suspensions were in the spherical form. This however has no effect upon the volumes. The volumes are therefore approximately correct.

With the exception of the reading for the horse cells, the viscosities of the suspensions increase with the cell volume. The relation is shown in the graph attached and is to be expected from theoretical considerations.

GRAPH SHOWING RELATIONSHIP of VOLUME of CELL to ABSOLUTE VISCOSITY.



The viscosity of the suspension of sheep cells is however somewhat greater than that of the horse, although the volume is less. This is unexpected and at the present time inexplicable to us.

With regard to the number of cells and size of cells, the general result is that as the size of the cells increases, so does the viscosity increase. This increase in viscosity is also noticeable with increase in number of cells within the limits discussed above.

In connection with the influence of the size of cell on viscosity, two important observations must be dealt with.

(1) Effect of carbon dioxide. Carbon dioxide is stated by Burton-Opitz to increase the viscosity of the blood whether administered to an animal or passed through the blood in vitro. This effect is unexplained in each case.

We shall consider the latter case first.

(a) Suspensions of washed cells.

A 16% suspension of washed ox cells was prepared in saline; the viscosity measured and thereafter CO_2 was passed through the suspension for five minutes. The CO_2 was obtained from a cylinder.

At/

At the end of this time the viscosity was again measured, the precaution being taken to flush out the viscosimeter with CO_2. It is true that CO_2 will tend to leave the blood and pass into equilibrium with the air, but, as we are concerned with any amount of CO_2 and not any specific amount of CO_2, this is a matter of no importance.

In a succession of three experiments, the following results were obtained:-

		Before.	After.
Ox	(1)	.0172	.0174
	(2)	.0169	.0174
	(3)	.0171	.0172

It will be observed that there is no significant difference in the viscosity before and after the passage of CO_2.

On examination of the suspensions under the microscope we observed that the cells were present in the spherical form which results from suspension of red cells in saline.

When the cells are in the spherical form it is impossible for them to change in shape as the result of the action of CO_2 and also very difficult for them to change appreciably in volume.

Moreover, there being no buffering salts present in the saline suspension, the CO_2 merely passes into solution and therefore must have an entirely different effect/

effect to that which it must have when passed into the blood.

(b) The same experiment using whole defibrinated ox blood.

CO_2 was again passed through the blood at a slow rate, to avoid excessive frothing and the viscosity measured, before and after.

The following are the results:-

		Before CO_2	After CO_2
Ox	(1)	.0432	.0436
	(2)	.0434	.0437
	(3)	.0431	.0438

Again it will be noted that the differences are so small as to be scarcely significant. The deduction to be drawn from this is:-

(a) that the viscosity of the plasma is not altered by the passage of CO_2 and this is in agreement with Burton-Opitz.

(b) that the viscosity of the corpuscular contents is not altered.

(c) that the volume of the corpuscular phase remains the same.

This is in agreement with the findings of Joffre and Poulton and with the very elaborate investigation of Dryerre, Millar and Ponder, in which it has been shown that the size of the cells does not alter even in the/

the presence of CO_2 tensions up to 80 mm.

By in vitro experiments we were unable to show that CO_2 has any effect on the viscosity of the blood.

The result obtained by Burton-Opitz was obtained in vivo and is open to a very simple explanation. It has been shown by Barcroft that the result of the administration of CO_2 is to cause the spleen to pour out into the blood-stream a large number of red-cells, with the result that the red-cell content may increase from 10% to 20%.

Such an increase in the volume of the corpuscular phase will of course account for the increase in viscosity and at the same time for the absence of any effect of CO_2 in vitro, where the corpuscular content is quite fixed.

(2) The effect of hypotonic saline.

One of the easiest methods of altering cell volume is by means of hypotonic saline. The blood of an animal, man, was selected and a 16% suspension made up in isotonic (.8%) NaCl. The cells are necessarily in the spherical form.

Equal vols. of this suspension were then mixed with solutions of hypotonic saline, so that the resulting 8% suspensions were in .8%, .7%, .6%, .5% and .4% NaCl. Solutions more hypotonic were found to produce haemolysis of some of the cells.

The/

The viscosities of the 8% suspensions were then measured in the usual way.

Tonicity.	Rel. Visc.	Abs. Visc.
.8	2.32	.0209
.7	2.37	.0213
.6	2.29	.0206
.5	2.27	.0204
.4	2.30	.0207

It will be seen that within the range of experimental error there is little change in the viscosity. This is rather surprising for we should expect the increase in the size of the cells to increase the viscosity, but we must remember that the passage of water into the cells decreases the viscosity of the cell contents, which factor plays an important part in the total viscosity.

The addition of water to the saline causes very little difference in its viscosity, for the viscosity of saline is very near that of water.

SECTION V. In this section we shall deal with the effect of the shape of the red-cell as a factor in the viscosity of blood.

It is well-known that red-cells can exist in two normal forms (1) Discoidal form in which they exist in/

in the blood-stream. (2) Spherical form such as they possess in isotonic saline.

In the discoidal form the diameter of human calls is 8.8 and in the spherical form 5.6 μ. In each case the volume is the same, 110 μ^3. In the case of the cells of any other animal the ratio

$$\frac{\text{diameter of spherical form}}{\text{diameter of discoidal form}}$$

will be the same, i.e. $\frac{5.6}{8.8}$.

We have already pointed out in the theoretical considerations that the form will have an effect on the viscosity, for the cells in the flat discoidal state are in the most stable position, therefore in the best position to resist deformation.

In order to investigate the effect of form on viscosity we have carried out the following experiment.

A suspension of cells is prepared in .85% NaCl such that the strength of the suspension is 16% and its viscosity measured.

Another suspension of cells of identical strength is prepared in .85% NaCl with the addition of 1% Ammonium Chloride. The effect of the addition of Ammonium Chloride is to render the cells of the suspension permanently discoidal. The viscosity of this 16% suspension is also measured.

Cells.	Form.	η
Human	discoidal	.0209
	spherical	.0203
Ox	discoidal	.0184
	spherical	.0171
Sheep	discoidal	.0156
	spherical	.0149

In each case the viscosity of the suspension of spherical cells was very slightly less than that of the suspension containing the cells in the discoidal form. The difference is however not very marked.

SECTION VI. Effect of Rouleaux Formation.

In defibrinated blood as normally used there is a great deal of rouleaux formation, the cells being collected together in long columns of twenty or more. Accordingly investigations were carried out to determine the effect of these phenomena on viscosity.

(1) Agglutination.

Agglutination is the aggregation of cells in masses of roughly spherical form. These masses may attain large size and be quite readily visible to the naked eye.

As a means of agglutination the tox.-albumin Ricin was used.

1 cc./

1 cc. of a 1 in 10,000 dilution of Ricin is quite sufficient to agglutinate 10 cc. of cell suspension in a few minutes, in the incubator at 37°C. This quantity of ricin has a negligible effect on the viscosity of saline.

Measurement of the viscosity of a 16% suspension of cells before and after agglutination

		Before.	After.
Man	(1)	.0214	.0120
	(2)	.0208	.0113
Ox	(1)	.0183	.0102
	(2)	.0186	.0105
	(3)	.0173	.0099

We find from the above experiments that the suspensions after agglutination show a much lower viscosity than before.

This is presumably due to the cells being collected into comparatively few large masses, instead of a large number of smaller masses.

The flow of the fluid is interfered with to a much lesser extent by the first condition than by the second.

(2) Rouleaux Formation.

The only known method of producing true rouleaux formation experimentally is that described by Sellards which is adapted to our purpose as follows.

A/

A sample of defibrinated blood is taken and divided into two portions. The viscosity of one portion is measured. A second portion is treated in order to produce rouleaux formation. The cells are first centrifugalised down, the supernatant serum is pipetted off and heated to 56°C. for 15 to 30 minutes as in the inactivation of complement.

The serum is then added to the red cells from which it is removed and the whole shaken up. Marked rouleaux occurs within 15 minutes. The viscosity of the sample in which the rouleaux are present is then determined and compared with that of the other sample.

The following results show the effect of rouleaux formation.

		1st sample.	2nd sample.
Ox	(1)	.0182	.0164
	(2)	.0176	.0159
Sheep	(1)	.0153	.0134
	(2)	.0146	.0129
Horse	(1)	.0121	.0109
	(2)	.0118	.0106

In four cases out of the six the effect of marked rouleaux formation was to diminish the viscosity but not to a very great extent. In one case the viscosity was lessened markedly as if agglutination had occurred and in the sixth case a slight increase in viscosity took place.

The/

The influence of rouleaux formation can be attributed to three factors as follows:-

(1) The cells tend to be present in large clumps and thus lessen the viscosity.

(2) Being in long cylinders they cannot lie near the wall of the tube.

(3) Rouleaux are always colliding with one another which produces the effect of a disorderly flow in the fluid which means an increased viscosity is observed.

The next section concerns a matter which cannot be adequately treated under any of the previous sections.

SECTION VII. Effect of haemolysis on viscosity.

This subject was first investigated by Burton-Opitz and Dettermann. The particular form of haemolysis which they considered was haemolysis by freezing and thawing. Burton-Opitz found that the viscosity of laked blood was less than normal. Dettermann on the other hand found that it was more viscous and attributed this result to the red cells containing highly viscous substances which were responsible for the greater viscosity of laked blood. Burton-Opitz continuing the investigation found that the essential point was, that if the cell envelopes were allowed to deposit out or were centrifugalised out, /

out, the viscosity of the laked blood becomes less, whereas if these were left, the viscosity became greater.

We performed experiments to decide the effect of the following factors:-

(1) Effect of laking by freezing and thawing the blood.

(2) Effect of laking by addition of saponin.

(3) Effect of laking by means of complement and immune body.

(1) To lake the blood by freezing and thawing was performed as described in a previous experiment by freezing the blood in a freezing mixture. When thoroughly frozen, the tube containing the blood was plunged into warm water. This process was repeated three times, by which time all the cells were disintegrated.

The viscosity of blood laked in this manner was invariably found to be greater than that of the unhaemolysed blood. The following results are examples of this.

		before.	after.
Ox	(1)	.0486	.0558
	(2)	.0472	.0512
Sheep	(1)	.0405	.0492
	(2)	.0423	.0501
Horse	(1)	.0452	.0512
	(2)	.0441	.0509

This therefore confirms Dettermann's observations.

The increase of viscosity appears to be due to the uniform distribution of haemoglobin, for, since the cells occupy 30% of the volume of the blood, and since the haemoglobin in them is in 30% concentration, the concentration of haemoglobin liberated on complete laking is no less than 10%. This amount of protein added to the 6% of serum proteins already present is sufficient to account for the high viscosity of the blood.

(2) Saponin.

The method adopted was to add solid saponin to the blood in quantities of 1 mgm. per cc. upwards.

Small quantities of saponin do not complete haemolysis but as larger quantities are used, complete haemolysis may be obtained.

In these cases in which incomplete haemolysis resulted, the degree of haemolysis was roughly estimated.

The following are results at 35°C.

Saponin/

Saponin.	Amount haemolysis.	η
.5 mgms per 1 cc of blood	0	.0426
1.0	tr.	.0433
2.0	M	.0451
3.0	V M	.0472
4.0	C	.0493
5.0	C	.0499
6.0	C	.0512
7.0	C	.0518
8.0	C	.0524
9.0	C	.0529
10.0	C	.0533
12.0	C	.0541
14.0	C	.0552
16.0	C	.0559
18.0	C	.0567
20.0	C	.0575

From this experiment we draw the following conclusions:-

(1) Viscosity increases steadily from no haemolysis to complete haemolysis.

(2) Even after complete haemolysis further additions of saponin cause increase of viscosity.

The first effect is easily explained on the same grounds as those given by Dettermann. The second effect however requires further investigation.

(3) Effect of complement and immune body.

In the case of complement amboceptor reactions the smallest amount of complement which will cause lysis of a given quantity of sensitized cells must first be determined.

A 6% suspension of Ox red cells was made up in .85% saline. To 100 cc. of this was added 1.5 cc. of immune body prepared by immunising a rabbit with increasing doses of ox cells. The cell suspension and immune body was then allowed to stand at room temperature for one hour.

A series of tubes were next set up and into each tube five cc. of the above sensitised ox cell suspension was placed.

To each tube was next added varying amounts of complement starting with .04 cc. and rising by .04 to .2 cc.

Complete lysis was found to take place with 0.1 cc of complement.

The viscosity of the laked blood in this tube was then determined.

The same experiment was performed with sheep cells and sheep immune body, and the viscosity of the laked blood determined. The results were as follows:-

Ox./

		Before.	After.
Ox	(1)	.0477	.0523
	(2)	.0469	.0506
Sheep	(1)	.0412	.0498
	(2)	.0418	.0506

It will be seen from these results that the viscosity is greater after laking of the cells. This result can scarcely be due to the addition of the very small amounts of immune body or complement which are present. In the case of complement the concentration is only 2% and of immune body, 1.5%. The large increase in viscosity must necessarily again be due to the increased distribution of haemoglobin in the fluid.

MATHEMATICAL CONSIDERATIONS.

While it is possible to apply mathematical treatment to the experimental results obtained with any one component of the total blood, it is quite impossible to do so with the whole blood itself. Although this is the case there are many theoretical points which are of interest both in connection with results and methods.

A most excellent treatment of the subject is to be found in "La Viscosité" by Brillouin.

Poiseuille's law can be expressed as follows:-

$$I = K \frac{P.D^4}{l}$$

where I is total outflow

K is a constant for the liquid

l is the length of the tube of diameter D.

and P is the difference of the pressure at the two ends of the tube.

The coefficient K is related to the viscosity coefficient η in a manner which may be deduced from the following theoretical considerations.

Take the case of a circular tube for which we must calculate the outflow from the whole cross-section of the/

the tube. The flow across a strip contained between two circles of radius r and $r + dr$ is $u \cdot 2\pi r \cdot dr$. The total outflow is then

$$I = \int_0^{r_2} u \cdot 2\pi r \cdot dr = \frac{2\pi P}{4\pi l} \int_0^{r_2} (r_1^2 - r^2) r \cdot dr$$

r_2 being the radius of the tube and equal to r when there is no slip at the wall.

Continuing

$$I = \frac{2\pi P}{4\eta l} r_1^2 \left(\frac{r_1^2}{2} - \frac{r_1^2}{4}\right)$$

which becomes

$$I = \frac{\pi P}{8\eta l} (r_1^4)$$

or substituting the diameter for the radius

$$I = \frac{\pi P}{8\eta l} \left(\frac{D^4}{16}\right)$$

whence

$$K = \frac{\pi}{128\eta}$$

K and η are thus entirely independent of the pressure, length or diameter of the tube and if values of K for pure distilled water can be found we can always find η in absolute units.

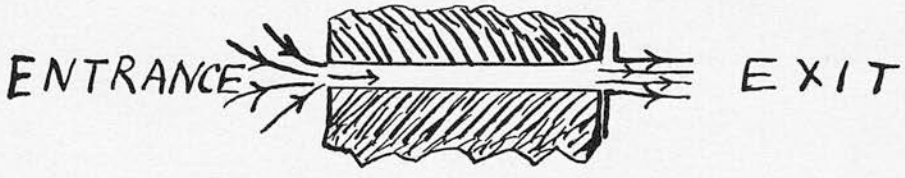
These values have been determined by Thorpe and Roger, Poiseuille, Slotte, Sprung, Stephan, Traube, and Rosenkrantz. The observations agree remarkably and give the following value for K at different temperatures.

constant.

the height of the effective column of fluid varies from the beginning of the experiment until its end because the upper level of the fluid is continually falling and the lower level continually rising, but its true value has been shown to be the mean of the levels at the beginning and at the end of the experiment. The value of C is small, it depends upon the diameter of the capillary and on the construction of the instrument as well as on the surface tension of the fluid. It is impossible to evaluate it theoretically and scarcely necessary to do so practically.

There are two further considerations of importance each of which limit Poiseuille's law to some extent.

(1) In the neighbourhood of the ends of the tube through which flow takes place, the lines of flow cease to be parallel to the axis of the tube because at these points the tube widens out into bulbs.



As /

As will be seen from the diagram the lines of flow at the entrance of the tube are different from those at the exit.

The consideration of the manner in which this affects the viscosity is a very difficult matter, but the final result may be given. The effect is to transform Poiseuille's expression into

$$\eta = \frac{P\pi r_1^4 - \rho I^2}{8\pi I(l+\lambda)}$$

There are thus two correction terms required. The one λ supplies the correction for the currents at the entrance but is clearly of no importance, if l is great compared to r, a condition which is satisfied in the Ostwald instrument.

The other term is proportional to the square of the outflow and only becomes of importance when the outflow becomes great. In the use of the Ostwald instrument it is however necessary that the outflow should not be great and hence the correction terms are not required in practice.

(2) The second limitation to Poiseuille's law also depends upon the quantity of fluid discharged in unit time, but for another reason. If the flow within the tube becomes very rapid, it may exceed what is termed the critical velocity. When the fluid enters the tube it does so at a certain pressure, and when it leaves/

leaves it, it does so at a smaller pressure for energy has been transformed into heat during the flow.
The difference of pressure ΔP is proportional to the rate of flow when that rate is small, but for high rates of flow it is proportional to the square of the rate. There is a certain value at which the simple proportionality ceases to occur and this value or rate of flow is called the critical velocity. It has been found that below the critical velocity the flow in the tube is orderly, while above the critical velocity the motion is turbulent. Now any velocity is proportional to some power of the density ρ, the viscosity η and the radius of the tube r that is

$$V_c = k \rho^x \eta^y r^z$$

where V_c is the critical velocity.
The dimensions of a density are M/L^3, those of a viscosity M/LT and the dimensions of a radius are L. So we have

$$\frac{L}{T} = \left(\frac{M}{L^3}\right)^x \left(\frac{M}{LT}\right)^y L^z$$

The coefficients of M give $x + y = 0$, the coefficients of T give $y = 1$ & $x = -1$ and the coefficients of L give $z = -1$ hence

$$V_c = \frac{k \eta}{\rho r}$$

that is, the critical velocity is a function of viscosity and is inversely proportional to the radius of the/

the tube.

For water k equals very nearly a thousand and so we have

$$V_c = \frac{1000\eta}{\rho r}$$

In the case of blood flowing through a tube of 1 mm. bore and of a density practically unity the critical velocity is thus 450 cm. per sec. Under the conditions of the Ostwald viscosimeter orderly flow will therefore always occur, for this velocity never occurs. In the case of even smaller capillaries as in the body, turbulent motion will occur even less readily, for the velocity of the blood never reaches the speed of 4.5 metres per sec.

The foregoing considerations have their bearing principally upon matters of technique and upon the use of the Ostwald viscosimeter. We have now to consider certain mathematical expressions which have been suggested as valuable in the description of experimental results.

These are:-

(1) Einstein's expression.

(2) Expression of Hatschek.

(3) Expression of Arrhenius.

all relating viscosity with volume of disperse phase.

(1) Einstein's expression deduced for a suspension of small rigid spheres separated widely in proportion to/

to their radius is

$$\eta = \eta_0 (1 + k \varphi)$$

Where η_0 is the viscosity of the dispersion medium η that of the suspension φ is ratio of volume of solid to total solid and k a constant. This is a linear expression, the viscosity increasing with the concentration. Einstein gives the value 2.5 for k . Hatschek gives 4.5 and Bancelin gives 2.9. This expression does not hold very well in practice and is applicable only to suspensoids containing small concentrations of solid phase. It obviously cannot apply completely to the viscosity of blood suspensions where $\eta = 5$ approx. and where η_0 is not greater than 1.5 for, if it were applicable φ would require to be greater than 1 which is impossible.

Recently it has been suggested that the relation

$$\eta = \frac{\eta_0 (1 + k \varphi)}{1 + \varphi^4}$$

can be used as a purely empirical expression for the viscosity of suspensoids and emulsoids. The practical and theoretical results are said to correspond to a remarkable degree, but the expression has no advantage over any other expirical formula. It will be noted that this expression takes no account of the size of the individual dispersed spheres.

(2)/

(2) Hatschek's formula. This expression is

$$\eta = \eta_0 \left\{ \frac{\sqrt[3]{P}}{\sqrt[3]{P} - 1} \right\}$$

the symbols having the same meaning as above.

The curve given by this expression is hyperbolic in appearance η increasing rapidly as φ approaches unity, that is, as the disperse phase increases in volume. This expression depends, from a theoretical point of view on the system having a certain "structure" which is supposed to be polyhedral. It has not been found to be applicable with any degree of accuracy to the viscosity of blood, although Murray-Lyon thinks that it is useful.

(3) Arrhenius has shown that the expression

$$\log \eta = kC.$$

can be used for expressing the viscosity of many sols. The expression is purely empirical and C itself is a "molecular concentration" which can scarcely be determined. The formula fails for many sols, k becoming negative (Ostwald).

It is not surprising that, as the conditions in simple organic sols are such as to defy mathematical expression, the conditions found in such a complex system as that of blood should yield little better result. Indeed, any attempt to combine into one expression the effect of many independent factors can only be regarded as unsound.

SUMMARY OF EXPERIMENTAL SECTION.

1. A series of viscosity determinations of oxalated plasma of the rabbit, ox, horse, guinea-pig and goat, sheep, rat, pig and man are given. Speaking generally the viscosity of such plasma is about 1.5 times that of water.

2. The viscosity of the serum of the same animals is given. It is about 10% less than that of the corresponding plasma. The explanation of this fact lies in the removal of fibrinogen.

3. The effect of various anticoagulants on the viscosity of plasma was investigated. Small quantities of oxalate up to 15 mgms. per 5 cc. have no effect. Larger quantities increase the viscosity considerably.

4. Sodium Citrate influences the results similarly to oxalate. We have not been able to determine the effect of Hirudin.

5. The effect of diluting serum with NaCl is to diminish its viscosity, the diminution occurring very rapidly in the lower degrees of dilution.

6. No constant results were obtained on heating serum to 56°C. In some cases the viscosity was slightly increased, in others slightly diminished.

7. An endeavour was made to obtain the viscosity of the corpuscular contents by separating the cell envelopes from blood laked by freezing and thawing. The experiments indicate that the viscosity of the contents is about 4.5 times that of water. The viscosity of a 15% Haemoglobin solution was found to be 2.7 times that of water. The high result for corpuscular contents is therefore not surprising.

8. An attempt was made to find the viscosity of suspensions of red cell envelopes in concentration equal to that which occurs in blood. The result shows that the viscosity of such a suspension is about 1.5 times that of water.

9. The number of cells present in a suspension exercises a very important effect on the viscosity. Determinations made in the case of the horse, rat, pig, rabbit, guinea-pig, ox and man show that it is quite useless to attempt to obtain consistent results in dilute suspensions. When stronger suspensions are investigated (from 32% to 4%) it is found that the viscosity of a suspension increases with the cell content comparatively slowly until about 20% of cells are present. Thereafter a great increase occurs.

10. From measurements of the viscosity of suspensions of equal numbers of cells of sheep, horse, ox, guinea-pig and man, it is concluded that as the size/

size of the cell increases so does the viscosity increase.

11. The passage of CO_2 through a suspension of red-cells does not appreciably alter the viscosity.

12. The passage of CO_2 through defibrinated ox-blood does not significantly change the viscosity.

13. Cell suspensions in saline of tonicity from .8% to .4% possess the same viscosity.

14. Suspensions of red-cells in the discoidal form are a little more viscous than suspensions of cells in the spherical form.

15. The effect of agglutination by ricin is to greatly diminish the viscosity of the suspension.

16. The effect of the marked formation of rouleaux is to diminish the viscosity of the blood, in most cases. Occasionally an increase occurs.

17. Laking of the blood by freezing and thawing increases the viscosity slightly. Laking by saponin increases the viscosity steadily until complete laking occurs. Further additions cause a marked increase in viscosity.

18. In the case of haemolysis by complement and amboceptor an increase of viscosity is shown to occur.

19./

19. A brief consideration of the applicability of some of the suggested mathematical formulae is added.

GENERAL SUMMARY.

Regarding the blood as a complex physical system it is quite impossible to find any universally valid explanation for its properties of viscosity, and, while it is impossible to reduce the expression of this property to any one general mathematical formula, the difficulty in dealing with the entire system arises from the fact that there are present many variables, few of which can alter independently of the others, and most of which it is impossible to measure with any degree of accuracy. e.g. Defibrination alters the viscosity of the plasma without altering the cells. Changes in the shape of the cells alters the viscosity of the blood as a whole but does not affect that of the plasma.

As soon as we split up the problem of the viscosity of the blood into the problems of its various components, the matter simplifies itself immediately. The only general statement which can be made, is that every substance or state which brings about a resistance to deformation plays its part in determining the final viscosity. If we could measure each one of these resistances in absolute terms, the viscosity of the entire system would be the sum of the whole. As it is, we can only show in a very approximate manner the extent to which each constituent of the blood influences the viscosity.

The plasma or serum possesses a viscosity slightly greater than that of water, and therefore plays a comparatively unimportant part in the viscosity of the blood.

So far as the simple investigation which we have carried out, shows, the resistance to deformation offered by the red-cell envelopes is likewise small and these also influence the total viscosity very little. The sum of these two effects however, would account for a viscosity about twice that of water.

By far the greatest contribution to the total viscosity appears to be made by the corpuscular contents, which are very viscid, their viscosity being nearly five times that of water.

The problem of the viscosity of the blood is thus similar to the problem of the viscosity of oil-droplets in water, provided that we imagine the oil-droplets to be surrounded by delicate envelopes of some substance such as soap. In each case the viscosity is dependent on the viscosity of the disperse phase; the oil, or the corpuscular contents, as the case may be.

This being the case, the quantity of the disperse phase must play a very unimportant part. It is to be noted that the volume of the disperse phase, (cells), can be increased in two ways,

(a)/

(a) by increasing the number of cells per cu. mm. of blood.

(b) by decreasing the amount of water in the plasma.

It is by one of these two methods that the viscosity of the whole blood appears to be affected under physiological conditions.

In connection with the importance of the number of cells we may recall the results obtained by Burton-Opitz on measuring the viscosity of the blood of an animal rendered anaemic by means of Phenyl Hydrazine and the increase of viscosity observed by many investigators after asphyxia.

The first of these results is explained by the much lower red-cell content giving rise to a decreased viscosity just as the viscosity is increased when the blood is diluted with normal saline. This explanation is even more clearly applicable to Burton-Opitz's experiments on anaemia produced by haemorrhage.
The effect of CO_2 on the viscosity of the blood appears principally due to the fact that in vivo the red-cell numbers increase; the effect is absent in vitro, for no such increase is possible.

In addition to this increase in the number of cells, the addition of CO_2 to the blood, in vivo brings about a redistribution of the water:-

(a)/

(a) between the plasma and cells

(b) between the plasma and tissue fluids.

These redistributions doubtless affect the problem, as also may the possible effect of large quantities of CO_2 on the viscosity of the corpuscular contents.

A most interesting piece of evidence in connection with this, is the fact observed by Burton-Opitz, that venous blood, although containing considerably more CO_2 than arterial blood, has an almost identical viscosity. This bears out the suggestion that the increase of viscosity obtained in vivo by the administration of CO_2 is not due to the effect of the CO_2 on the volume of the cell, on the viscosity of the contents, or on the viscosity of the plasma. The only remaining factor is the number of cells.

Changes in the water-content of the blood, i.e. the viscosity of the plasma occur even more frequently in vivo. Burton-Opitz found that the injection of water greatly decreases the viscosity as might be expected, and in general that the viscosity of the blood increases with an increase of specific gravity. As an example of this fact we can recall his experiments with hot and cold water baths and the rather unexpected result, that hot-air baths render the blood more viscous, whereas hot-water baths render it less viscous. The explanation of this in terms of the water content is simple, for hot-air baths are known to concentrate the plasma because/

because of the loss of water from the skin, whereas hot-water baths do not exercise this effect.

Finally we may consider the effect to which estimations of the viscosity of the blood may be held to be of physiological or clinical importance. All observers are agreed (see especially Murray-Lyon and Albutt) that there is no condition in which an estimation of blood-viscosity has been found to be of value in diagnosis, prognosis, or treatment. The principal reason for this is the fact that the viscosity of the blood, unless it varied outside very wide limits, cannot have any important effect on the organism as a whole, for such variations are immediately compensated for, by a mechanism which can readily undergo changes to a far greater extent than can the viscosity.

The work done by the heart in moving the blood round the vessels is,

$$W = P.\ M.$$

where P is the mean pressure in the arterioles and M is the mass of blood expelled in the time over which the work is measured.

This mass is dependent upon D, the density of the blood, R, the rate of the heart beat and O the ventricular output. The pressure developed in the arteries is a measure of the peripheral resistance to outflow and this depends upon the mean capillary diam-

eter/

diameter and the viscosity of the blood, as well as, of course, on the force exerted by the heart.

So we have

$$W = f(M) = f(D, R, O)$$

and also

$$W = f(P) = f(\eta \cdot d)$$

This will give some idea of the complexity which occurs and which is added to, by the facts that by Starling's law of the heart

$$R = f(O)$$

and by Burton-Opitz's observations

$$\eta = f(D)$$

The result of all this interdependence of the various factors, is that we can compensate for any alteration in η in several ways, some of which must be continually called into play in the living animal.

E.g. Should the viscosity of the blood rise 10% we will get one of the following results.

(a) A diminution of pressure if W and d remain constant.

(b) An increase in the capillary diameter d, if the pressure P and the work of the heart W are to be constant.

(c)/

(c) An increase in W, the work of the heart, if P and d are to be constant.

Any slight change of viscosity can therefore be immediately compensated for in several ways.

An excellent example of this is shown in the sequence of events after haemorrhage. The blood-volume is rapidly made up by passage of fluid from the tissues and η falls. If the arterioles are unaltered in diameter P the pressure, falls to the same percentage extent, but in fact (McDowall 1927) a vaso-constriction takes place, P is restored to normal, and the work of the heart remains the same. The diminution in viscosity has therefore no effect in lessening the work done although it would have had such an effect, if the compensating vaso-constriction had not intervened.

It will be seen from the above, that, unless we can accurately determine d, the mean diameter of the capillaries, we must entirely ignore measurements of blood viscosity in vivo and be guided by the blood-pressure .

Although the problem of the viscosity of the blood is interesting from a physical point of view, it is necessarily one which can have very little interest when considered from the point of view of the intact animal, for its effects are much too dependent on the pressure or absence of other compensating effects which are known to occur with greatest readiness.

Indeed, any attempt to ascertain the importance of the viscosity of the blood in the intact animal is exactly analogous to the determination of the viscosity of an unknown fluid driven by an unknown and variable force, through a capillary of unknown and variable bore.

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